Conformation and dynamics of a single DNA molecule confined by walls(Poster session 2, New Frontiers in Colloidal Physics : A Bridge between Micro- and Macroscopic Concepts in Soft Matter)

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高分子を壁の間にはさんで擬似２次元空間内に拘束すると、自由空間とは大きく異なった運動を示す。本研究では、２枚のガラス板の間にDNAを挟み、その間隔を変えながらDNA １分子の運動を蛍光顕微鏡を用いて観察した。取得した画像を二値化してＤＮＡの重心位置の並進拡散と長軸の回転拡散を解析した。

Introduction

A movement of polymer in a quasi two-dimensional space is greatly different from that in a free space. In order to obtain dynamical properties of the DNA in the two-dimensionally confined space, we used single DNA molecules placed between two glass plates. Translational diffusion and rotational relaxation of the single DNA molecules were experimentally measured from direct observation of the fluorescently labeled DNA.

Experiment

T4GT7 DNA, about 166 kbp or 56 μm in contour length, was solved in TE buffer solution (5 mM Tris-HCl, 0.5 mM EDTA) containing 4 % (v/v) 2-mercaptoethanol, 4.6 mg/ml glucose, 0.2 mg/ml glucose oxidase, and 0.036 mg/ml catalase. Major groove binding fluoroscent dye, YOYO-1, was added as a probe of DNA. We put the DNA solution in the glass cell controlled to several μm thickness and observed a single DNA molecule. Fluorescent images were obtained with video camera whose sampling rate was 1/30, and sequential images were recorded in PC as a movie data.

Result

Figure 1 shows a typical picture of a single DNA molecule. The DNA internally fluctuates and translationally diffuses inside the glass cell. First, the fluorescent images are converted into binarized images. We assume the center of mass of the binarized DNA image as that of the

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DNA, and then we track the DNA in the sequential images. Mean square displacement (MSD) of DNA is estimated from the position $\mathbf{R}(t)$ of the center of mass as,

$$\mathrm{MSD} = < (\mathbf{R}(\mathrm{t}+\Delta\mathrm{t}) - \mathbf{R}(\mathrm{t}))^2 > .$$

Figure 2 shows a logarithmic plot of MSD under 2 μm in solution thickness. Total tracking time is 152 sec. The slope of the MSD corresponding to diffusion coefficient exhibits normal diffusion ($\sim t^1$) for 10 sec and below lag time. On the other hand, we estimate the rotational relaxation time τ_r of the same DNA molecule from the time correlation function of the unit vector $\mathbf{u}(t)$ parallel to the long axis diameter as shown in Figure 1 right. We define the long axis vector of the DNA in place of an end-to-end vector. The calculated correlation function is fitted by an exponential curve with single relaxation time as,

$$< \mathbf{u}(t+\Delta t) \cdot \mathbf{u}(t) >= \exp(-\Delta t/\tau_r).$$

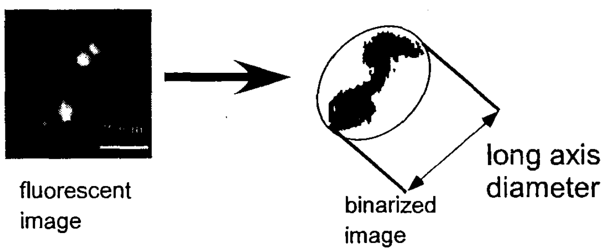

Figure 1: Fluorescent image of a single DNA molecule (left handed panel), and binarized image of the DNA (right handed panel).

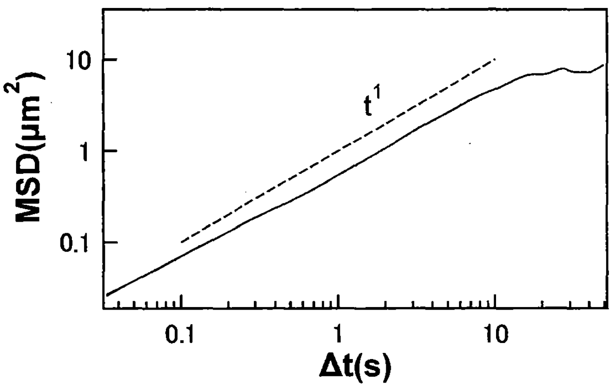

Figure 2: MSD of a single T4GT7 DNA molecule under 2 μm in sample spacing.

References

[1] M. Matsumoto et al, J. Polym. Sci. **30** (1992), 779.